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NUTRITIVE VALUE OF RAPESEED MEAL FOR POULTRY

by



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The undersigned certify that they have read, and recommend to the Faculty of Graduate Studies and Research for acceptance, a thesis entitled "Nutritive value of rapeseed meal for poultry" submitted by Prakash Chandra C. Seth, B. Sc., B.V.Sc. and A.H., M.V.Sc., in partial fulfilment of the requirements for the Degree of Master of Science.

ABSTRACT

Three experiments were conducted to study various aspects of the nutritive value of rapeseed meal for poultry.

In the first experiment three samples of commercial rapeseed meal prepared from *B. campestris*, *B. napus* and Bronowski type rapeseed were each separated into low hull and high hull fractions by a process called air classification. The composition of the meals and their fractions was determined by usual laboratory procedures and the metabolizable energy values of meals and their fraction were determined with chicks. The results obtained indicated that the air classification procedure was reasonably effective in separating rapeseed meal into two fractions: one low in fibre and high in protein and energy content; the other high in fibre and low in protein and energy content. Average values for the metabolizable energy content of the meals and their low hull and high hull fractions were 1,610, 2,016 and 1,287 kcal/kg. Chemical composition data showed a higher concentration of glucosinolates and tannins in the low hull fractions than in the high hull fractions suggesting that these deleterious factors tend to be more concentrated in the endosperm than in the pericarp of the seed.

In the second experiment the solubility in saline and acid solutions of the nitrogen in samples of rapeseed meal collected at hourly intervals over a period of 72 hours in a prepress-solvent processing plant was studied. Highly significant differences in the solubility of the nitrogen in

different samples were found. On the basis of the results obtained it would seem worthwhile to investigate the causes of the solubility differences noted.

In the third experiment the effect of including 20% rapeseed meal in the ration of broiler-type chickens on the incidence of perosis and the manganese requirement of broiler-type chickens was studied. Inclusion of rapeseed meal in the ration was found to significantly increase the incidence of perosis, however, no evidence was obtained in this study to indicate that the manganese requirement of the chickens was increased by the inclusion of rapeseed meal in their diet.

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INTRODUCTION

Rapeseed production in Western Canada has increased rapidly in recent years. A particularly spectacular increase in production occurred during the period 1969 to 1971 when production of rapeseed rose from 37,000,000 bushels to 98,500,000 bushels. The increased interest in rapeseed production has been partly the result of the Government of Canada's program to encourage Canadian farmers to grow less wheat and partly because rape is a cash crop that is well adapted to production in Western Canada. Concurrent with the increase in rapeseed production interest has developed in the processing of rapeseed for oil and meal, the former being used extensively in cooking oil, salad oil and margarine, the latter in livestock and poultry rations.

The use of rapeseed meal in rations for livestock and poultry has been plagued with difficulties, one of which has been the low metabolizable energy content of rapeseed meal in relation to that of other common vegetable protein supplements such as soybean meal. Many studies have been conducted to determine why rapeseed meal is inferior in this respect and suggestions have been made concerning how the energy value of rapeseed meal may be improved. One of these suggestions has involved the possibility of separating rapeseed meal into low fibre and high fibre fractions. The thinking behind this suggestion was that perhaps by conducting such a separation one might obtain a low fibre, high energy product suitable for feeding monogastric animals and birds

and a high fibre fraction adapted to ruminant feeding.

The work reported herein is concerned with a study of the nutritive value of rapeseed meal and of fractions derived therefrom by air classification. In addition, it is concerned with variations in the solubility of the nitrogen in rapeseed meal and with the effect of feeding rapeseed meal to broiler-type chickens on the incidence of perosis.

PART I

COMPOSITION AND METABOLIZABLE ENERGY VALUE OF
RAPESEED MEAL AND OF FRACTIONS DERIVED THEREFROM

Review of Literature

Composition of Rapeseed Meal

Wetter (1955) reported on the potential volatile isothiocyanate content of *Brassica napus* and *Brassica campestris* type rapeseed and showed that there was little difference between the two types of seed. However, in a later publication, Wetter (1957) found a considerable difference between the potential oxazolidinethione content of these two kinds of rapeseed, the *B. napus* type was found to be much higher than the *B. campestris* type. Clandinin *et al.* (1959) found that the potential amounts of oxazolidinethione in rapeseed were dependent on the variety of seed and the environmental conditions under which the seed was grown.

Numerous reports have appeared on the proximate composition, amino acid, mineral and vitamin content of rapeseed meal. Klain *et al.* (1956) reported values for choline, niacin, pantothenic acid, riboflavin and thiamine for two samples of rapeseed meal. Karvanek *et al.* (1964) presented data on the total phosphorus, phytin phosphorus, copper, iron, manganese and nickel content of rapeseed and rapeseed meal. Clandinin (1967) reported on the proximate composition, amino acid, calcium and phosphorus content of expeller, prepress-solvent and solvent-processed rapeseed meals. Work on the

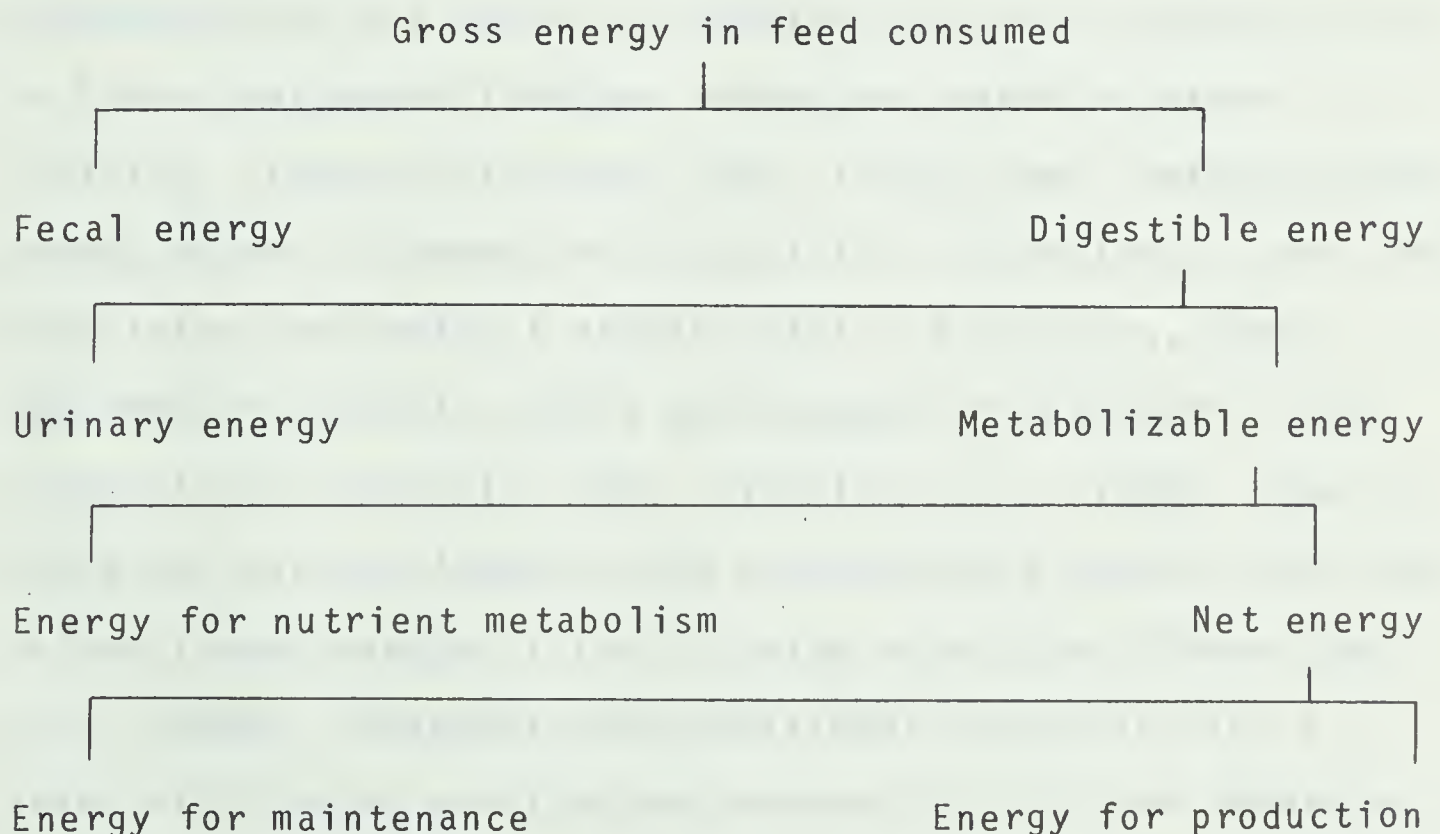
selenium content of rapeseed meal has been reported by Arthur (1971).

A comprehensive review on the composition of rapeseed meal has recently been prepared by Clandinin *et al.* (1972).

Metabolizable Energy Value of Rapeseed Meal for Poultry

1. Partition of energy in feeds

Various parameters have been used to evaluate the caloric content of feedstuffs eg. digestible energy, metabolizable energy, net energy etc. The relationship between same is shown diagrammatically below.



Net energy appears to be the most scientific measure of feed energy, since it takes into account all energy losses due to digestion and metabolism and thus measures the actual

amount of energy available for useful work (Fraps and Carlyle, 1942). However, its determination is very tedious and the values obtained, even for replicates, are quite variable. This is because net energy is influenced by the level of feed intake, the balance of nutrients in the ration, the type of animal used in the determination and the environmental conditions to which the animals are subjected during the determination (Hill and Anderson, 1958).

On the other hand, metabolizable energy measures the total energy of the feed that is available for the metabolic purposes of the body. It is the gross energy of the feed minus the energy lost in the feces and urine. Metabolizable energy determinations are readily conducted with avian species since in birds undigested feed and urine are voided together. In addition, studies have shown that, in the fowl, metabolizable energy values of feeds are essentially unaffected by level of feed intake and rate of growth (Hill and Anderson, 1958), egg production (Hill, 1965) and balance of nutrients in the ration (Carew and Hill, 1961; Sibbald *et al.*, 1962). Age of the bird, has no effect on the determination except where the metabolizable energy of fat is being determined (Renner and Hill, 1960b). Moreover, the additional correction for a state of nitrogen equilibrium proposed by Hill and Anderson (1958) to correct for variations in the protein deposited due to differences in feeding schedule has brought about even greater agreement in metabolizable energy values reported by

different workers for the same feedstuff.

2. Determination of metabolizable energy

The determination of metabolizable energy using chickens has been carried out in several laboratories (Fraps and Carlyle, 1942; Carpenter and Clegg, 1956; Hill and Anderson, 1958; Matterson *et al.*, 1958; Potter and Matterson, 1960; Sibbald *et al.*, 1960; Sibbald and Slinger, 1963a).

In one procedure for determining metabolizable energy, (Matterson *et al.*, 1958) test material is fed either alone or in combination with adequate amount of minerals and vitamins. This method has a great drawback in that the ration contains either inadequate or excessive amounts of several nutrients and its utilization may be abnormal, particularly when fed over prolonged periods of time.

A second procedure which has been used extensively by Hill and Anderson (1958) and by Potter and Matterson (1960) consists of feeding a basal ration containing a chemically pure reference ingredient of known metabolizable energy content and a test ration. The test ration is similar to the basal ration except a portion of the reference material in the basal ration is replaced by the test material, the metabolizable energy of which is under determination. The metabolizable energy content of the basal ration and the test ration are determined and the metabolizable energy content of the material under test is calculated. The method assumes a constant metabolizable energy value for the reference

ingredient. The method may be criticized because the test rations may contain excessive amounts of several nutrients.

A third procedure for the determination of metabolizable energy is that of Sibbald *et al.* (1960) and Sibbald and Slinger (1963a). It consists of feeding a practical-type control ration and a test ration formulated by replacing a part of the control ration with the ingredient to be assayed. The metabolizable energy contents of the two rations are determined and then the metabolizable energy of the ingredient under test is calculated. This method is subject to criticism on the grounds that the test ration may not be balanced nutritionally.

3. Metabolizable energy value of rapeseed meal for poultry

Sibbald and Slinger (1963a) using chicks, reported that the metabolizable energy of one sample of solvent-processed rapeseed meal which contained 43.1% protein (Nx6.25) was 1,670 kcal per kilogram (dry-matter basis). On the other hand Sell (1966), using hens, obtained a value of 2,290 kcal per kilogram (dry-matter basis) for one sample of solvent-processed rapeseed meal which contained 38.3% protein.

Using the analytical procedure of Hill and Anderson (1958) Lodhi *et al.* (1969) reported that the average metabolizable energy content of nine samples of prepress-solvent and solvent-processed rapeseed meal was 1,203, 1,313 and 1,782 kcal per kilogram (dry-matter basis), respectively, for 4 week old chicks, 6 week old chicks and hens fed rations

containing 30 per cent rapeseed meal for at least 21 days. Subsequently Rao and Clandinin (1970) confirmed the fact that, age of chickens used in the determination affected the metabolizable energy values obtained for rapeseed meal. In addition, these workers showed that the method used for the determination of the metabolizable energy content of rapeseed meal significantly affected the values obtained. Average values for two samples of rapeseed meal using the methods of Hill and Anderson (1958) and of Sibbald and Slinger (1963a) were 1,245 and 1,654 kcal per kilogram (dry-matter basis) respectively.

In a recent review, based on the published data referred to above and on unpublished data obtained at The University of Alberta and the University of Guelph, Clandinin *et al.* (1972) recommended that for poultry a metabolizable energy value of 1,760 kcal per kilogram (air-dry basis) be used for rapeseed meal. These authors state that when this value is used in poultry ration formulation feed conversion does not appear to be affected adversely.

4. Factors affecting the metabolizable energy value of rapeseed meal for poultry

(a) Presence of goitrogen(s) in rapeseed meal

Numerous workers (Turner, 1946; Blakely and Anderson, 1949; Witz *et al.*, 1950; Frölich, 1953; Dow and Allen, 1954; Klain *et al.*, 1956 and Clandinin *et al.*, 1959) have reported

thyroid enlargement as a result of feeding rapeseed meal to poultry. In general, meal produced from *Brassica napus* type seed has been shown (Klain *et al.*, 1956 and Clandinin *et al.*, 1959) to cause a greater degree of thyroid enlargement than meal produced from *Brassica campestris* type seed. This is considered to be due to the higher (-)-5-vinyl-2-oxazolidinethione content (Astwood *et al.*, 1949a, 1949b and Carroll, 1949 isolated goitrin from rapeseed and identified it as L-5-vinyl-2-thiooxazolidone which has more recently been called (-)-5-vinyl-2-oxazolidinethione, Greer, 1962) of rapeseed meal produced from *B. napus* type seed as compared to that produced from *B. campestris* type seed (Wetter and Craig, 1959; Clandinin and Bayly, 1960).

(-)-5-vinyl-2-oxazolidinethione (goitrin) occurs in rapeseed as a glucosinolate (progoitrin). In the presence of the enzyme(s) myrosinase, which is also present in rapeseed, progoitrin is hydrolysed into glucose, bisulphate and 2-hydroxy-3 butenyl isothiocyanate (Greer, 1956 and Van Etten *et al.*, 1969). The latter compound is unstable and gives rise to the cyclic antithyroid compound (-)-5-vinyl-2-oxazolidinethione (Kjaer, 1960).

In Canada, during the commercial extraction of oil from rapeseed, myrosinase present in the seed is destroyed by heat (Youngs, 1965) and as a consequence, little, if any, goitrin is found in Canadian rapeseed meal. However, if an exogenous source of myrosinase, such as ground mustard seed, finds its way into feeds containing rapeseed meal some

goitrin may be released (Lodhi *et al.*, 1970). In addition, it has been shown that a small amount of the progoitrin in a feed may be converted to goitrin by intrinsic myrosinase-like enzymes produced by intestinal microorganisms (Gmelin and Virtanen, 1959). It is probably because of the presence of exogenous and intrinsic myrosinase that the thyroid enlargement referred to in the first paragraph of this section occurs.

In a series of experiments designed to study the effects of rapeseed meal, progoitrin and synthetic goitrin (± 5 -vinyl-2-oxazolidinethione) on the uptake and release of radio-iodine from the thyroid glands, Clandinin *et al.*, (1966) observed that initially iodine uptake by the thyroid glands and iodine released by the glands was decreased. However, after the chicks had any one of these supplements for three or four weeks, uptake of radio-iodine by the hypertrophied glands was greatly increased and daily secretion of radio-iodine from the latter glands was found to be normal. These workers concluded that chicks fed rapeseed meal, progoitrin or goitrin, like rats fed rapeseed (Hercus and Purves, 1936; Kennedy and Purves, 1941; Griesbach, 1941; Griesbach *et al.*, 1941; Purves, 1943; Griesbach and Purves, 1943) eventually reach physiological equilibrium at increased thyroid-to-body weight ratios. Matsumoto *et al.*, (1968), using progoitrin, confirmed the findings of Clandinin *et al.* (1966).

Scow and Foglia (1951), found that thyroidectomy resulted in slower absorption of glucose. Increased rate of absorption of glucose and galactose, dextrose and oleic acid was reported by Althausen and Stockholm (1938), after administration of thyroxine. Several workers (Fink, 1944; Russell and Nasset, 1953; Sun *et al.*, 1954) have reported that thyroidectomy or hypothyroidism in monogastric animals results in depression in the amounts of gastric and intestinal secretion.

Lodhi *et al.* (1970) have conducted studies to determine whether the low metabolizable energy value of rapeseed meal was, in part, due to the presence in rapeseed meal of glucosinolates or products derived from same by myrosinase hydrolysis. These workers found that the goitrin and isothiocyanates released by adding ground mustard seed, as a source of myrosinase, to rations containing 30 per cent of rapeseed meal had no effect on the metabolizable energy value of the rations. In addition these workers reported that adding a relatively high level (0.045%) of ± 5 -vinyl-2-oxazolidinethione to a soybean meal-type ration had no effect on its metabolizable energy value.

(b) Available carbohydrates in rapeseed meal

Hrdlicka *et al.* (1965) reported that the soluble sugar, starch and pentosan contents of rapeseed meal were 3.2, 5.8 and 10 per cent respectively. In a literature review Hardinge *et al.* (1965) reported that soybean meal contained

2.0, 9.0, 1.8, 2.4 and 5.0 per cent, respectively, of reducing sugars, sucrose, dextrin, starch and pentosans.

Bolton (1957) reported that the available carbohydrate content of soybean meal (49% protein) was 17.6 per cent. Available carbohydrate in two samples of rapeseed meal and one sample of soybean meal (50% protein) was determined using a chemical method and a chick bioassay by Lodhi *et al.* (1969). Results of the chemical assay indicated that the total soluble sugars and starch in rapeseed meal and soybean meal (50% protein) were 15.0 and 23.6% respectively. The bioassay showed that the available carbohydrate in the samples of rapeseed meal and soybean meal were 6.9 and 14.1% respectively. The difference in amount of available carbohydrate in rapeseed meal and soybean meal was found to account for 290 kcal per kilogram of the total difference in the metabolizable energy value of these two feedstuffs.

(c) Available protein in rapeseed meal

Information on the absorability of nitrogen in rapeseed meal for poultry is limited. Kubota and Morimoto (1965), using surgically altered hens, found that the absorbability of the nitrogen in one sample of rapeseed meal was 72.6%, where rapeseed served as the sole source of protein. More recently, Drouliscos and Bowland (1969) reported apparent and true nitrogen absorbability for rapeseed meal was 74 and 79% respectively for weanling rats when the meal served as the sole source of protein. The corresponding

values obtained for mature rats were 79 and 83% respectively.

Lodhi *et al.* (1970) measured the apparent absorbability of nitrogen from rapeseed meal and soybean meal (50% protein). Results based on the determination of unabsorbed nitrogen in the terminal fifth of the small intestine of six week old chicks fed rations in which either rapeseed meal or soybean meal served as the sole source of protein showed that the absorbability of the nitrogen in rapeseed meal and soybean meal was 79.9 and 85.4% respectively. When the data on protein absorbability was used to calculate the contributions which protein in rapeseed meal and soybean meal make to metabolizable energy, the lower absorbability of the nitrogen in rapeseed meal together with its lower protein content accounted for 711 kcal per kilogram of the difference in metabolizable energy value of the two feedstuffs when rapeseed meal served as the sole source of protein.

(d) Presence of tannins in rapeseed meal

Clandinin and Heard (1968) showed that rapeseed meal contains about 3% tannin and speculated that on the basis of the work of Vohra *et al.* (1966) that rapeseed meal may contain sufficient tannins to adversely affect the metabolizable energy value of rations which contain high levels of rapeseed meal.

More recently, Yapar and Clandinin (1972) reported that the removal of tannins from rapeseed meal significantly increased the metabolizable energy value of rapeseed meal for

chicks. Furthermore, addition of tannins extracted from rapeseed meal to a ration based on soybean meal resulted in a reduction in its metabolizable energy value.

(e) Level of rapeseed meal fed

Lodhi *et al.* (1970) presented data which indicated that the level of rapeseed meal in the test ration affected the metabolizable energy value obtained. This finding was confirmed by Yapar and Clandinin (1972) who obtained metabolizable energy values of 1,237 and 1,761 kcal per kilogram when rapeseed meal was incorporated in the test ration at the 30% level and as the sole source of protein respectively.

(f) Age of chickens fed rapeseed meal

Lodhi *et al.* (1969) presented data which indicated that the metabolizable energy value of rapeseed meal for the chicken increases as the age of chicken increases. This finding has been confirmed by Rao and Clandinin (1970) and Bayley *et al.* (1970).

(g) Length of time chickens fed rapeseed meal

Lodhi *et al.* (1969) showed that metabolizable energy values for rapeseed meal tended to be higher if the chickens and hens were fed rapeseed meal containing rations for about three weeks rather than for only five days before fecal collections for metabolizable energy determinations were made. Yapar and Clandinin (1972) noted a similar effect.

(h) Fat content of rapeseed meal

The fat content of rapeseed meal varies depending on the process used in the extraction of oil. Generally speaking solvent and prepress-solvent-processed meals contain 1 to 2% fat while expeller-processed meals of high protein quality run 6 to 8% fat (Clandinin and Tajcnar, 1961; Clandinin, 1967). Bayly *et al.* (1970) have shown that the metabolizable energy value of rapeseed meal is correlated with the fat content of the meal. Hence, other things being equal, the metabolizable energy value of expeller-processed rapeseed meal may be expected to be higher than that of solvent or prepress-solvent-processed rapeseed meals.

(i) Fibre content of rapeseed meal

Rapeseed meal contains 12 to 13% crude fibre, Clandinin (1967). Carpenter and Clegg, (1956) have reported a high negative correlation between the crude fibre content of a feedstuff and its metabolizable energy value. Hence it seems reasonable to suggest that any process that would reduce the crude fibre content of rapeseed meal should tend to increase its metabolizable energy value for poultry.

Studies at The University of Alberta

The purpose of this work was to study the composition and metabolizable energy value of rapeseed meal and of high hull and low hull fractions derived therefrom with a view to determining whether or not a low hull fraction would be more suitable for feeding poultry than regular rapeseed meal.

Experimental

Three samples of commercial prepress-solvent-processed rapeseed meal, one prepared from *Brassica campestris* type rapeseed, another from *Brassica napus* type seed and a third from a variety of seed known as Bronowski served as the starting material for this study. A large quantity of each sample of commercial rapeseed meal was reground to pass through a 200 mesh screen and separated by air classification¹ into two fractions, one supposedly of low hull content and the other of high hull content.

The commercial meals (hereafter called regular meals) and their fractions were subjected to chemical analyses. Moisture, crude protein (Nx6.25), crude fibre and tannin contents were determined by methods described by the Association of Official Agricultural Chemists (1965). Potential levels of oxazolidinethione and volatile isothiocyanates were determined by methods developed by Astwood (1949) and Wetter (1955 and 1957), respectively. Amino acid analyses were

¹Work was done at Bauer Brothers Company, Springfield, Ohio, U.S.A.

conducted on acid hydrolysates of the regular meals and their fractions with a Technicon Amino Acid Analyzer. Determination of calcium was done by ethylenediaminetetraacetic acid (EDTA) titration (Vogel, 1961) with the modification that 2 ml of 20% sucrose solution was added to 10 ml aliquots of sample before titrating with EDTA. Sucrose was added to keep the calcium in solution when the magnesium was precipitated by potassium hydroxide. Estimation of phosphorus was carried out by the metavanadate method of Jackson (1958) with the following modifications. The sample (1 gram) was first soaked for 30 minutes in 5 ml of a solution containing magnesium nitrate (950 g per l) to which approximately 20 mg of magnesium oxide was also added. The sample was next dried and ashed in a muffle furnace at 800°C until free of carbon. The ash was dissolved in 15 ml of concentrated HCl and filtered. The balance of the procedure was as described by Jackson (1958).

The metabolizable energy values of the regular rapeseed meals and their fractions were determined by the method of Sibbald and Slinger (1963a). The basal diet used is shown in Table 1. The test material was incorporated at the 30% level (moisture-free basis) as a replacement for an equal percentage of the test diet. To 100 parts of the experimental diets 3 parts of mineral and vitamin mixtures were added and correction for this was made when the calculations of metabolizable energy were done.

TABLE 1. - Composition of basal diet

Ingredients	%
Ground corn	35.0
Ground wheat	35.0
Ground barley	17.0
Dehydrated alfalfa meal	4.0
Dried whey	4.0
Stabilized tallow	5.0
Mineral mixture	+ ¹
Vitamin mixture	+ ²

¹Supplied in milligrams per 100 grams ration: CaCO_3 , 1000; Ca_2HPO_4 , 1040; NaCl , 240; $\text{MnSO}_4 \cdot \text{H}_2\text{O}$, 13; ZnCO_3 , 6.5; Cr_2O_3 , 300.

²Supplied per 100 gram ration: riboflavin, 0.22 mg; calcium pantothenate, 0.44 mg; niacin, 1.11 mg; menadione sodium bisulfite, 0.23 mg; vitamin B_{12} , 1.0 μg ; choline chloride, 22.77 mg; vitamin A, 500 I.U.; vitamin D_3 , 801 I.C.U.; DL methionine, 50 mg.

For determining metabolizable energy, male cross-bred (Dominant White males x White Plymouth Rock females) chicks were used. The chicks were housed in electrically heated, temperature controlled battery brooders, with raised wire screen floors, in a well ventilated, temperature controlled room. Electric light was provided on a continuous basis in the brooder room. During the first 28 days the chicks were fed a balanced starter and water on an *ad libitum* basis. At 28 days of age, selection and randomization of chicks was done by the method described by Sibbald and Slinger (1963a) in order to obtain a low level of variation in the derived metabolizable energy data. Each experimental ration was fed to quadruplicate groups of 10 chicks from 28 to 42 days of age.

Excreta were collected at 24 hour intervals at 40, 41 and 42 days of age. The three-day collections from each group were pooled and kept at -18°C until analyzed. Incorporation of chromic oxide in the rations at the 0.3% level eliminated the need for quantitative measurement of feed intake and excreta. The methods of processing excreta and of conducting chemical analyses for moisture, nitrogen, combustible energy and chromic oxide have been described by Hill and Anderson, (1958); Hill *et al.*, (1960). Metabolizable energy was computed in accordance with the method described by Sibbald and Slinger (1963a).

Results and Discussion

Summarized in Table 2 are data on the metabolizable energy values for the regular rapeseed meals and their low hull and high hull fractions. Analyses of variance and application of Duncan's multiple range test (Steel and Torrie, 1960) showed that there were highly significant within variety differences ($P < 0.01$) in the metabolizable energy content of regular meal, low hull and high hull fractions prepared from *B. napus* and Bronowski rapeseed and significant differences ($P < 0.05$) in the metabolizable energy content of regular meal, low hull and high hull fractions from *B. campestris* rapeseed.

Shown in Table 3 are data on the crude protein and crude fibre contents of the regular rapeseed meals and their low hull and high hull fractions. Analyses of variance and application of Duncan's multiple range test (Steel and Torrie, 1960) showed that there were highly significant within variety differences ($P < 0.01$) in the crude protein and crude fibre contents of regular meal, low hull and high hull fractions. These differences are generally reflected in the metabolizable energy values of the regular meals and their low hull and high hull fractions.

Considerable variation was observed (Table 3) in the effectiveness of the separation of the three regular meals into high protein-low fibre and low protein-high fibre fractions. For example, considerably more fibre was left in the low hull *B. campestris* fraction than in the low hull

TABLE 2. - Metabolizable energy value of rapeseed meals and of fractions derived therefrom

Type or variety of seed from which meal produced	Metabolizable energy, 6 weeks, kcal/kg ¹		
	Regular	Low hull	High hull
<i>Brassica campestris</i>	1,678 ^b	1,490 ^a	1,477 ^a
<i>Brassica napus</i>	1,354 ^a	2,091 ^b	755 ^c
Bronowski	<u>1,799^a</u>	<u>2,467^b</u>	<u>1,630^c</u>
Averages	1,610	2,016	1,287

¹ Values represent averages for quadruplicate groups and are based on moisture-free meal. Within a meal type, values without a common letter in their superscripts are significantly different, $P < 0.05$ for *B. campestris*, $P < 0.01$ for *B. napus* and Bronowski.

TABLE 3. - Chemical composition¹ of rapeseed meals and of fractions derived therefrom

Type or variety	<i>B. campestris</i>			<i>B. napus</i>			Bronowski		
	Regular	Low hull	High hull	Regular	Low hull	High hull	Regular	Low hull	High hull
Crude protein (Nx6.25), %	39.2 ^a	42.06 ^b	34.58 ^c	39.66 ^a	44.08 ^b	31.11 ^c	44.74 ^a	50.2 ^b	36.54 ^c
Crude fibre, %	17.44 ^a	10.30 ^b	22.96 ^c	17.17 ^a	7.76 ^b	24.89 ^c	15.29 ^a	8.12 ^b	22.54 ^c
Calcium, %	0.84 ^a	0.69 ^b	0.96 ^c	0.64 ^a	0.44 ^b	0.83 ^c	0.70 ^a	0.50 ^b	0.89 ^c
Phosphorus, %	1.33 ^a	1.39 ^a	1.02 ^b	1.30 ^a	1.44 ^a	0.90 ^b	1.08 ^a	1.24 ^b	0.86 ^c
Isothiocyanate, gm/kg	5.06 ^a	5.96 ^a	4.49 ^b	5.0 ^a	4.80 ^a	3.75 ^b	0.51	0.40	0.33
Oxazolidinethione, gm/kg	2.74 ^a	2.6 ^a	1.84 ^b	8.67 ^a	8.88 ^a	6.52 ^b	0.10 ^a	0.40 ^b	0.10 ^a
Tannins, %	4.26 ^a	3.63 ^b	2.47 ^c	4.65 ^a	3.78 ^b	2.0 ^c	3.61 ^a	3.87 ^a	2.43 ^b

¹ Values are based on moisture-free meal. Within a variety, values without a common letter in their superscripts are significantly different (P<0.05).

B. napus fraction. Such differences are not considered to be related to the type of seed from which the regular meal was produced but rather to physical differences in the reground meals which affected the air classification of the regular meals into low fibre and high fibre fractions. This area requires further study.

One problem was encountered with the fractions. It was noted that when the fractions were included in rations at high levels there was a tendency for the chicks to develop beak necrosis. Since beak necrosis is caused by feeding rations that are too finely ground (Conklin and Maw, 1930), the problem could probably be avoided by pelleting and crumbling the fractions following air classification in order to change them from powdery to granular texture. This, of course, is a common practice that is used to improve the texture of feeds for poultry.

Carpenter and Clegg (1956) showed that there was a high negative correlation between the crude fibre content of a feedstuff and its metabolizable energy value. The lower metabolizable energy value of rapeseed meal compared to that of soybean meal has been attributed to its lower crude protein content, lower digestibility of its protein, lower amount of available carbohydrate and higher crude fibre content (Lodhi, 1970) and perhaps, in part, to its high content of tannins (Yapar and Clandinin, 1972). The results obtained in this study demonstrate quite clearly that it is possible by air

classification to separate regular rapeseed meal into two fractions: one, high in protein and low in fibre; the other, low in protein and high in fibre. The results also suggest that the first mentioned fraction is likely to be about 25% higher in metabolizable energy value than regular rapeseed meal while the second mentioned fraction is likely to be about 25% lower in metabolizable energy value content than regular rapeseed meal.

Also summarized in Table 3 are data on the tannin content of regular meals, low hull and high hull fractions prepared from *B. campestris*, *B. napus* and Bronowski rapeseed. Analyses of variance and application of Duncan's multiple range test (Steel and Torrie, 1960) showed that there were highly significant within variety differences ($P < 0.01$) in the tannin content of regular meal, low hull and high hull fractions, except in the case of meals produced from Bronowski rapeseed where the differences in the tannin content of the regular meal or the low hull fraction and the high hull fraction were significant ($P < 0.01$). The tannin contents of the regular rapeseed meals in this study were in close agreement with the values reported by Yapar and Clandinin (1972).

The fact that the tannin contents of the low hull fractions were always higher than the tannin contents of their respective high hull fractions (Table 3) indicates that the extractable tannins tend to be concentrated in the endosperm of the seed rather than in the pericarp. Furthermore, if, as

has been suggested by Yapar and Clandinin (1972), the tannins in rapeseed meal tend to lower the metabolizable energy value of rapeseed meal, any increase in the tannin content of the high protein-low hull fraction would tend to adversely affect its metabolizable energy value. It would therefore appear that solution to the tannin problem rests not with air classification but on some, as yet undetermined, tannin extraction process or in the hands of the plant breeder.

Summarized in Table 3 are data on the oxazolidinethione and isothiocyanate contents of regular meals, low hull and high hull fractions produced from *B. campestris*, *B. napus* and Bronowski rapeseed. Analyses of variance and application of Duncan's multiple range test (Steel and Torrie, 1960) showed that there were significant within variety differences ($P < 0.05$) between the high hull fractions and the regular meals or low hull fractions prepared from *B. napus* and *B. campestris* rapeseed. Rapeseed meal produced from the Bronowski variety had a very low oxazolidinethione content. With respect to isothiocyanates, there were significant differences ($P < 0.05$) in the isothiocyanate contents of the high hull fraction and low hull fraction or regular rapeseed meal produced from *B. campestris* rapeseed. Similarly in the case of the *B. napus* variety there were highly significant differences ($P < 0.01$) between the isothiocyanate contents of the high hull fraction and the low hull fraction or regular meal. In the case of meals from the Bronowski variety the isothiocyanate contents

of regular meal, low hull and high hull fractions were not significantly different. In view of the fact that the low hull fractions produced from types of seed that contained appreciable amounts of oxazolidinethione and isothiocyanates were higher in these deleterious substances than the corresponding high hull fractions, it would appear that these substances are concentrated more in the endosperm than in the pericarp of the seed. As previously stated it appears that the oxazolidinethione and isothiocyanate contents of the low hull fractions are higher than those of the regular meals or the high hull fractions. However, these substances have been shown to have no adverse effects on the metabolizable energy content of rapeseed meal for chickens (Lodhi *et al.*, 1970). Hence, the increased concentration of oxazolidinethione and isothiocyanates in the low hull fractions cannot be considered as affecting the metabolizable energy value of the low hull fractions. Values obtained for isothiocyanates and oxazolidinethione contents of the rapeseed meals used in this study were in good agreement with the values reported by Lodhi *et al.* (1969) and Rao and Clandinin (1970).

Also summarized in Table 3 are data on the phosphorus and calcium contents of the regular meals, the low hull and the high hull fractions produced from *B. campestris*, *B. napus* and Bronowski rapeseed. Analyses of variance and application of Duncan's multiple range test (Steel and Torrie, 1960)

showed that there were significant within variety differences ($P < 0.05$) between the calcium content of the regular meals, the low hull and the high hull fractions. Significant within variety differences ($P < 0.05$) were found in the phosphorus content between the high hull fractions and the low hull fractions or regular meals in case of meals from *B. campestris* and *B. napus* rapeseed. In the case of meals from the Bronowski variety of rapeseed there were significant within variety differences ($P < 0.05$) in the phosphorus content between the high hull fraction, the low hull fraction and regular meal. On critical examination of the calcium content of regular meal, low hull and high hull fractions it appears that calcium occurs more in the pericarp than in the endosperm of the seed. However the reverse appears to be true in case of the distribution of the phosphorus. The values for calcium and phosphorus content of rapeseed meal were in close agreement with the values reported by Clandinin (1967).

The amino acid composition of the three rapeseed meals and their low hull and high hull fractions are presented in Table 4. The amino acid values obtained on the regular rapeseed meals were in close agreement with those reported for rapeseed meal by Clandinin *et al.* (1972). Statistical analyses of the data did not show any consistent changes in amino acid composition resulting from air classification of the regular meals into low hull and high hull fractions. Perhaps if the separation into low hull and high hull fractions

TABLE 4. - Amino acid composition¹ of rapeseed meals and of fractions derived therefrom

Type or variety	<i>B. campestris</i>			<i>B. napus</i>			Bronowski		
Meal or meal fraction	Regular	Low hull	High hull	Regular	Low hull	High hull	Regular	Low hull	High hull
Aspartic acid ²	6.58 ^a	6.67 ^b	7.03 ^a	6.51 ^a	7.08 ^c	6.73 ^b	7.27 ^a	6.69 ^b	6.61 ^b
Threonine	4.26 ^b	4.37 ^{ab}	4.27 ^a	4.02	4.30	4.24	4.32 ^a	4.14 ^{ab}	4.02 ^b
Serine	4.24 ^a	4.30 ^c	4.36 ^b	4.17 ^a	4.25 ^b	4.23 ^a	4.45 ^{ac}	4.21 ^b	4.15 ^c
Glutamic acid	17.30 ^a	18.63 ^c	18.21 ^b	17.14 ^a	19.41 ^a	17.38 ^a	18.30 ^{ab}	19.82 ^a	17.46 ^b
Proline	6.50 ^a	6.48 ^b	6.21 ^b	6.13 ^{ab}	6.18 ^b	5.95 ^a	6.23 ^{ab}	6.42 ^a	6.05 ^b
Glycine	4.90 ^a	5.22 ^b	5.09 ^b	4.68 ^a	5.19 ^b	4.91 ^{ab}	4.98 ^a	5.15 ^a	4.82 ^b
Alanine	4.27	4.29	4.34	4.13 ^a	4.39 ^b	4.22 ^{ab}	4.61	4.58	4.48
Valine	4.81	5.04	4.91	4.63 ^a	5.20 ^b	4.97 ^{ab}	5.11	5.16	5.14
Cystine	2.39	2.09	2.44	2.12	2.17	2.29	3.20	3.35	3.10
Methionine	1.92 ^{ab}	1.93 ^b	1.91 ^a	1.70 ^a	1.81 ^b	1.72 ^b	1.86	1.94	1.83
Isoleucine	3.80 ^a	4.01 ^b	3.91 ^a	3.64 ^a	4.03 ^b	3.90 ^b	3.97	4.05	4.03
Leucine	6.65 ^a	7.14 ^b	7.01 ^a	6.52 ^a	7.03 ^b	6.76 ^b	7.18	7.17	7.08
Tyrosine	2.28 ^a	2.44 ^b	2.24 ^b	2.13 ^a	2.38 ^b	2.37 ^b	2.30	2.34	2.31
Phenylalanine	3.70 ^a	3.98 ^b	4.04 ^b	3.59 ^a	4.00 ^b	3.94 ^b	3.93	3.96	3.97
Ammonia	2.42 ^a	2.51 ^b	2.51 ^a	2.30 ^a	2.79 ^b	2.52 ^{ab}	2.32	2.48	2.61
Lysine	5.87 ^a	6.33 ^b	5.86 ^a	5.73 ^a	6.22 ^b	5.94 ^{ab}	5.90	5.96	5.74
Histidine	2.67 ^a	2.78 ^b	2.61 ^a	2.57	2.79 ^b	2.60 ^a	2.72	2.68	2.61
Arginine	5.61	5.81	5.67	5.47	5.99 ^b	5.73 ^a	6.07	6.20	5.88

¹ Within a variety values without superscripts are not significantly different. Within a variety values without a common letter in their superscripts are significantly different (P<0.05).

² Amino acid values are expressed as a percentage of the protein (Nx6.25).

had been more effective, differences in the amino acid patterns in the regular meals and their fractions would have been apparent. However, even if differences in amino acid patterns had been noted, it is unlikely that they would have had any effect on the metabolizable energy value of the meal fractions since Carew and Hill (1961) have shown that deficiencies or excesses of amino acids in a ration have no effect on the metabolizable energy value of the ration.

Summary

A study was made on metabolizable energy value and composition of rapeseed meal and of fractions derived therefrom by air classification.

Results showed that it was possible to separate rapeseed meal into two fractions, one of lower hull and higher protein content than regular rapeseed meal, the other of higher hull and lower protein content than regular rapeseed meal. The low hull fraction was found to contain about 25% more metabolizable energy than regular rapeseed meal while the high hull fraction was found to contain about 25% less metabolizable energy than regular rapeseed meal.

The result also showed that soluble tannins, oxazolidinethione, isothiocyanates and phosphorus appear to be concentrated in the endosperm rather than the pericarp of the seed while the reverse appears to be the case with respect to calcium.

No consistent changes in amino acid distribution

resulting from the air classification of regular rapeseed meal into low hull and high hull fractions was observed. It is possible that had a better separation of the rapeseed meals into low hull and high hull fractions been effected some consistent changes in the proportions of the various amino acids in the protein of the fraction might have been noted.

PART II

VARIATION IN THE SOLUBILITY OF NITROGEN IN PREPRESS-SOLVENT-
PROCESSED RAPESEED MEAL

Review of Literature

Almquist *et al.* (1935) suggested solubility tests for the estimation of protein quality. They proposed the Protein Quality Index (P.Q.I.) for rating protein-rich feedstuffs. It is a factor obtained from four chemically determined values which comprise the copper precipitable, hot water soluble, phosphotungstic acid precipitable and pepsin undigestible fractions of the total nitrogen. A close correlation was reported between the P.Q.Is. for various animal by-products and the growth of chicks. Similarly, significant correlations between the P.Q.Is. of soybean meals, cottonseed meals and pea meals and their respective Gross Protein Values (G.P.Vs.) have been reported by Evans and St. John (1945).

Lund and Sandstrom (1942) reported the separation of plant proteins into five fractions by successively extracting with water, 5% KCl solution, 70% ethyl alcohol at 70°C, and 0.2% potassium hydroxide, the residue forming the fifth fraction. Using this technique on soybean, cottonseed and pea meals, Evans and St. John (1945) reported a positive correlation between the 0.2% potassium hydroxide soluble nitrogen and the G.P.Vs. for the meals.

Barnes and Woodham (1963) described the separation

of nitrogen into saline (0.5M NaCl) soluble, acid (6N HCl) soluble and alkali (0.02N NaOH) soluble fractions. It was found that only the determination of percentage nitrogen solubility for ground-nut meals in 0.5M sodium chloride and for cottonseed meal in 0.02N sodium hydroxide showed promise for application as useful routine methods for assessing protein quality in these feedstuffs. In this work nitrogen solubilities were studied in relation to the G.P.Vs.

Hrdlicka *et al.* (1964) reported that the method of extraction of fat from rapeseed affects the water, saline and alkali soluble nitrogen fractions of rapeseed meal. It was found that rapeseed meal produced by the De Smet procedure was of the poorest quality. Olcott and Fontaine (1942) found that the higher the processing temperature of cottonseed meal the lower the solubility of the nitrogen in 3.0% sodium chloride solution. A correlation between saline soluble nitrogen and the growth of rats was found.

Anwar and Clandinin (1971) studied the solubility of nitrogen in rapeseed meals produced by the solvent, prepress-solvent and expeller-methods of processing in relation to their G.P.Vs. These workers found a positive correlation between nitrogen solubility in saline (0.5M NaCl) and acid (6N HCl) solutions and the G.P.Vs. of the meals. Nitrogen solubility in alkali (0.02N NaOH) appeared to be a less suitable method for estimating the G.P.V. of rapeseed meal.

Studies at The University of Alberta

The following investigation was undertaken to determine whether variation occurred in the solubility in saline and acid solutions of the nitrogen in rapeseed meals produced in a prepress-solvent oil seed processing plant.

Experimental

Seventy-two samples of commercial rapeseed meal collected at hourly intervals at a prepress-solvent-processing plant were used in the study. Each sample was ground to pass through a 20 mesh screen and thoroughly mixed to permit accurate sampling.

Total nitrogen was determined in duplicate on each meal by the Kjeldahl method. Nitrogen soluble in saline solution was determined by shaking duplicate 1 gram samples of each meal for 1 hour in 100 ml of 0.5M sodium chloride solution in an incubator room at 37°C., filtering the samples through #1 Whatman paper and determining the nitrogen content of the filtrate by the Kjeldahl method. Percentage nitrogen soluble in 0.5M sodium chloride solution was calculated in relation to the total nitrogen found in the sample of meal. Nitrogen soluble in 6N hydrochloric acid was determined in a similar manner.

Results and Discussion

Summarized in Table 5 are data on the saline and acid soluble nitrogen contents of the 72 samples of rapeseed meal collected at hourly intervals over a period of three days.

TABLE 5. - Nitrogen content¹ of rapeseed meals collected at hourly intervals and percentages of same soluble in saline and acid solutions

RSM no.	Total N, %	% N soluble in		RSM no.	Total N, %	% N soluble in	
		0.5M NaCl	6N HCl			0.5M NaCl	6N HCl
1	5.80	41.42	44.87	37	5.76	44.97	47.31
2	5.73	39.35	43.19	38	5.80	38.88	47.33
3	5.82	40.50	44.88	39	5.70	43.47	48.29
4	5.66	43.15	45.98	40	5.78	43.24	46.49
5	5.73	41.62	47.03	41	5.80	43.28	46.34
6	5.69	43.32	46.84	42	5.87	37.81	47.54
7	5.70	42.02	45.79	43	5.71	41.31	46.59
8	5.67	44.08	46.91	44	5.76	43.34	50.09
9	5.67	43.03	47.09	45	5.82	43.00	48.57
10	5.79	42.14	46.72	46	5.70	42.05	46.69
11	5.68	42.78	46.57	47	5.81	44.60	49.78
12	5.75	42.91	48.22	48	5.64	44.15	47.25
13	5.65	48.23	47.88	49	5.80	45.48	49.29
14	5.75	42.78	46.09	50	5.80	43.83	48.32
15	5.68	43.62	47.58	51	5.66	43.23	49.18
16	5.67	42.95	47.71	52	5.70	43.90	48.32
17	5.74	42.44	46.61	53	5.70	43.73	47.06
18	5.75	42.96	46.69	54	5.71	43.64	47.76
19	5.76	42.58	46.75	55	5.72	43.83	48.12
20	5.73	43.80	46.51	56	5.71	44.37	48.65
21	5.72	42.32	45.90	57	5.71	44.40	47.90
22	5.72	42.83	48.34	58	5.70	42.24	48.03
23	5.71	43.34	46.58	59	5.65	43.90	48.12
24	5.78	42.99	47.84	60	5.64	45.13	47.08
25	5.64	42.96	46.50	61	5.73	44.46	48.01
26	5.76	42.83	46.74	62	5.70	43.45	46.77
27	5.68	42.87	48.77	63	5.80	43.38	47.59
28	5.79	42.40	47.84	64	5.61	42.98	47.89
29	5.77	42.72	47.57	65	5.84	44.65	48.04
30	5.76	43.08	46.65	66	5.76	43.75	45.63
31	5.82	44.20	46.17	67	5.75	42.93	47.70
32	5.66	46.19	48.27	68	5.74	41.84	48.35
33	5.80	42.93	49.31	69	5.71	44.03	47.78
34	5.59	42.58	49.73	70	5.78	44.74	49.82
35	5.76	44.44	48.52	71	5.84	43.90	48.92
36	5.79	44.45	47.92	72	5.88	43.74	48.40
Averages					5.74	43.19	47.60

¹Values represent averages of duplicates.

Analyses of variance (Steel and Torrie, 1960) showed that there were highly significant differences ($P < 0.01$) in the saline soluble and the acid soluble nitrogen contents of the rapeseed meal samples. The differences in saline and acid soluble nitrogen between replicates within samples were small supporting the precision of the determinations. Significant positive correlations ($r = 0.273$ and $r = 0.496$ for saline and acid soluble nitrogen respectively) were found between saline and acid soluble nitrogen values and the time of collection of the samples of rapeseed meal. Higher solubility values were obtained toward the end of the collection period. It is unfortunate that the collection period was not longer. Had it been longer a change in the trend of solubilities might have been observed.

The saline and acid solubility values reported herein are lower than those reported by Anwar and Clandinin (1971). While the saline and acid concentrations used in this study were similar to those used by the above-mentioned workers, the time of hydrolysis was perhaps better controlled in this study and may account for the difference in values obtained. This opinion is supported by unpublished work of Rao (1970) in which it was found that nitrogen solubility increases with increasing time of exposure to saline or acid solution.

Hrdlicka *et al.* (1964) reported saline soluble nitrogen to range from 12.4 and 28.0 per cent for rapeseed

meals produced by various commercial oil seed processing methods. These values are lower than those found in this study. However these workers used about three times as much sodium chloride in their saline solution as was used in the present study.

Summary

A study was undertaken to determine whether variation occurred in the solubility in saline and acid solutions of the nitrogen in rapeseed meals produced at hourly intervals in a prepress-solvent oil seed processing plant.

Results showed that there were highly significant differences in the saline soluble and the acid soluble nitrogen contents of the rapeseed meal samples. Positive correlations were found between saline and acid soluble nitrogen values and the time of collection of the samples of rapeseed meal. No cyclic pattern in nitrogen solubilities however, was established in this study.

PART III

EFFECT OF INCLUDING RAPESEED MEAL IN THE RATION OF BROILER-TYPE CHICKENS ON THE INCIDENCE OF PEROSIS AND THE AVAILABILITY OF MANGANESE

Review of Literature

Holmes and Roberts (1963) reported that the inclusion of rapeseed meal at the 30% level in the diet of growing chickens caused an increased incidence of perosis. Since Clandinin and Heard (1968) have reported that rapeseed meal contains about 3% of tannins and tannins have been shown by Jurd and Geissman (1956) to form complexes with metal ions it seems reasonable to postulate that perhaps the higher incidence of perosis noted by Holmes and Roberts (1963) on rapeseed meal containing diets was due to a partial deficiency of manganese. Manganese, of course, has been shown by Wilgus *et al.* (1936) and Gallup and Norris (1939) to prevent perosis when included in the diet at an appropriate level.

Studies at The University of Alberta

The purpose of this work was to determine whether the inclusion of rapeseed meal in the ration of broiler-type chickens increased the incidence of perosis by decreasing the availability of manganese in the ration.

Experimental

Two isocaloric, isonitrogenous basal rations were formulated, Table 6. One of the basal rations contained soybean meal as the main source of supplementary protein while

TABLE 6. - Composition of basal rations

Ingredients	1	2
	%	%
Ground wheat	61.49	46.99
Soybean meal	30.00	19.50
Rapeseed meal	-	20.00
Dehydrated alfalfa meal	1.00	1.00
Stabilized tallow	3.00	8.00
Ground limestone	1.75	1.75
Calcium phosphate	1.50	1.50
Iodized salt	0.25	0.25
Zinc oxide	0.01	0.01
Vitamin premix	1.00 ¹	1.00 ¹

¹ Supplied per 100 gm ration: Vitamin A, 500 I.U.; vitamin D₃, 83.3 I.C.U.; vitamin E, 2.2 I.U.; menadione sodium bisulfite, 0.11 mg; riboflavin, 0.44 mg; calcium pantothenate, 1.1 mg; niacin, 2.2 mg; choline chloride, 18.5 mg; folic acid, 0.33 mg; vitamin B₁₂, 1.0 µg; methionine, 50.4 mg; and procaine penicillin, 1.1 mg.

the other contained 20% of rapeseed meal as a replacement for a part of the soybean meal contained in the first mentioned basal. Each basal ration was supplemented with 0, 1/8, 1/4 and 1/2 lb of manganese sulphate monohydrate per ton of ration to yield eight ration treatments.

Quadruplicate groups of twenty broiler-type chicks (Dominant White males x White Plymouth Rock females) of mixed sexes (1/2 male, 1/2 female) were placed on each ration. Feed and water were supplied *ad libitum*.

From hatching to four weeks of age the chicks were brooded in electrically heated thermostatically controlled battery brooders with raised screen floors in a temperature controlled laboratory. At four weeks of age the chicks were transferred to floor pens in a brooder house, the replicates on each treatment being placed in the same pen.

The chicks were weighed in groups at four weeks of age and the incidence and severity of perosis was recorded by treatment and sex when the chicks were six weeks old.

Results and Discussion

Summarized in Table 7 are data showing the incidence of perosis in chicks fed rations containing rapeseed meal and soybean with or without supplementary manganese. Analysis of variance (Steel and Torrie, 1960) showed that there was a significant increase ($P < 0.05$) in the incidence of perosis in the chicks fed rations containing rapeseed meal as compared to those fed rations containing soybean meal. The incidence

TABLE 7. - Incidence of perosis

Supple- mental manganese, p.p.m.	Sex ²	Basal 1 - SBM					Basal 2 - RSM				
		Severity of perosis ¹					Severity of perosis ¹				
		1	2	3	4	Total	1	2	3	4	Total
0.0	M	-	-	-	1	1	2	-	-	4	6
	F	-	-	-	1	1	-	-	1	-	1
18.4	M	-	1	-	-	1	-	1	2	1	4
	F	-	-	-	-	-	-	-	-	-	-
36.9	M	-	-	-	-	-	1	-	-	1	2
	F	-	-	-	-	-	-	-	-	-	-
73.8	M	2	1	-	-	3	1	1	-	2	4
	F	-	-	-	-	-	-	-	-	1	1
Totals						6 ^b	18 ^a				

¹Rated 1 to 4 in order of increasing severity of perosis. Values without a common letter in their superscripts are significantly different (P<0.05).

²M=Male, F=Female.

of perosis was found to be higher in the case of males than in the case of females. Although differences were noted in the incidence of perosis in the various groups fed different levels of manganese, no trend for decreasing incidence of perosis with increasing levels of supplementary manganese was apparent. Overall incidence of perosis was 3.85 and 0.89 per cent for the rapeseed meal and the soybean meal fed groups respectively, and 4.7, 1.45, 0.32 and 3.55 per cent for 0, 18.4, 36.9 and 73.8 p.p.m. of manganese supplementation, and 5.3 and 0.36 per cent for males and females respectively.

Results of this experiment show that the feeding of rapeseed meal at the 20 per cent level increases the incidence of perosis in growing chickens. These findings are in agreement with those reported by Holmes and Roberts (1963).

Rapeseed meal contains a high level of tannins (Clandinin and Heard, 1968). Since tannins have been shown to form metallic ion complexes (Jurd and Geissman, 1956) one might postulate that the tannin in rapeseed meal could cause the formation of such complexes and thus make the metallic ions less available nutritionally. If such were the case one would expect that the addition of extra minerals should alleviate the deficiency caused by the above suggested complexing. However, in the present experiment it was found that supplemental manganese did not lower the incidence of perosis hence the results obtained do not support the hypothesis.

If the results are considered in the light of the

findings of Briggs and Lillie (1946), one might suggest that higher incidence of perosis in the rapeseed meal fed groups as compared to the soybean meal fed groups was due to hypothyroidism caused by the presence of goitrogen in the rapeseed meal. These workers showed an increased incidence of perosis in chicks fed a diet containing 0.5 per cent thiouracil over a period of five weeks, and that the condition was not prevented by the addition of manganese, choline, niacin, biotin or riboflavin. These authors suggested that the thyroid glands exercise some control over the incidence of perosis. However, Holmes and Roberts (1963) have shown that the goitrogenic principles present in rapeseed meal seem to have no influence on the perotic syndrome. Hence, in view of these seemingly contradictory reports it would be inappropriate to conclude that the goitrogenic substances in rapeseed meal are responsible for the perosis noted in chicks fed high levels of rapeseed meal.

The higher incidence of perosis noted in males as compared to females could be due to the higher body weight in case of males as compared to females which causes more load on the leg joints and which, in turn, might tend to cause the Achilles tendon to slip from its condyles. On the other hand the faster rate of bone growth in male chickens as compared to that in female chickens may be related in some manner to the problem, Jull (1951).

Summary

A study was conducted to determine whether the inclusion of rapeseed meal in the ration of broiler-type chickens increased the incidence of perosis by decreasing the availability of manganese in the ration.

Results showed that when broiler-type chickens were fed rations in which soybean meal or a combination of rapeseed meal and soybean meal served as the supplementary sources of protein a significantly higher incidence of perosis was noted in the chicks fed the latter ration as compared to chicks fed the former. Increasing the level of manganese in the rapeseed meal containing ration did not appear to lower the incidence of perosis.

GENERAL DISCUSSION

Results of the studies reported herein would appear to indicate that it is possible by air classification to separate rapeseed meal into two fractions: one of higher protein and lower fibre content than regular rapeseed meal; the other of lower protein content and higher fibre content than regular rapeseed meal. The data obtained suggest that the former is likely to be about twenty-five per cent higher in metabolizable energy content than regular rapeseed meal while the latter is likely to be about twenty-five per cent lower in metabolizable energy than regular rapeseed meal. The higher protein-lower fibre fraction should, because of its higher energy content, be suitable for inclusion in rations for birds and monogastric animals which as a general rule require rations that are reasonably high in energy content. On the other hand, the lower protein-higher fibre fraction should be quite suitable for inclusion in rations for ruminant animal which are better adapted to handle more fibrous feeds than birds and monogastric animals.

Two problem areas were noted in the work on air classification of rapeseed meal into high protein-low fibre and low protein-high fibre fractions that warrant further study. The first relates to the fact that the effectiveness of separation of the three rapeseed meals into low fibre and high fibre fractions varied considerably. The separation obtained on the *B. campestris* type meal was quite inferior to

that obtained on the *B. napus* and Bronowski meals. Whether this was the result of differences in the oil extraction processes used to produce the meals or to variations in the conditions used in the air classification procedure is not known. A second problem encountered centered around the fineness of grind of the fractions derived from the air classification procedure. All fractions were sufficiently powdery as to constitute a dust hazard in a feed mixing mill. In addition, when incorporated in rations at relatively high levels they produced feeds that were of such fine texture as to cause beak necrosis when fed to young chicks. It would seem, however, that this problem could be avoided by pelleting and crumbling the fractions after air classification. This is a common practice used to improve the texture of poultry feeds.

Of fundamental interest was the finding that the tannins and glucosinolates in rapeseed tend to be concentrated in the low hull rather than the high hull fraction suggesting that same are concentrated in the endosperm rather than the pericarp of the seed. It would thus appear that if a high protein-low fibre fraction free of these deleterious factors is to be produced for feeding birds and monogastric animals a special extraction procedure for the removal of these deleterious factors will have to be developed or the plant breeder will have to produce varieties of rapeseed that are free of these factors. In the latter regard, obtaining

glucosinolate free varieties of suitable oil composition appears to be only a few years away, Downey (1972).

In the studies on the solubility in saline and acid solutions of the nitrogen in rapeseed meals it was found that considerable variability in solubility occurs. It would seem desirable in future studies to determine what processing variables (moisture content of seed, temperatures and times of cooking, conditioning, desolventizing and toasting) affect nitrogen solubility in the meal. In addition an extensive study should be made to determine what variation in nitrogen solubility can be tolerated without risk to the nutritive quality of the meal.

A limited amount of evidence was obtained in this study which suggests that broiler rations containing high levels of rapeseed meal tend to induce perosis in chicks. It was postulated that the increased incidence of perosis was due to decreased availability of manganese in the ration as a result of complexing between tannins in rapeseed meal and manganese in the ration. However, no evidence was obtained in the studies to support this contention. Further studies involving rations containing higher levels of rapeseed meal than was used in this study and involving other factors which might be related to the perosis syndrome are required before a satisfactory explanation of this problem is obtained.

BIBLIOGRAPHY

- Almquist, H. J., E. L. R. Stocksted and E. R. Halbrook, 1935. Supplementary values of animal protein concentrates in chick rations. *J. Nutrition*, 10: 193-211.
- Althausen, T. L., and M. Stockholm, 1938. Influence of the thyroid gland on absorption in the digestive tract. *Am. J. Physiol.* 123: 577-588.
- Anwar, A. and D. R. Clandinin, 1971. Nitrogen solubility as a means of estimating the gross protein value of rapeseed meal. *Poultry Sci.* 50: 135-136.
- Arthur, D., 1971. Selenium content of some feed ingredients available in Canada. *Can. J. Anim. Sci.* 51: 71-74.
- Association of Official Agricultural Chemist. 1965. *Official Methods of Analysis*, 10th ed. Washington, D. C.
- Astwood, E. B., M. A. Greer and M. G. Ettlinger, 1949a. The antithyroid factor of yellow turnip. *Science* 109: 631.
- Astwood, E. B., M. A. Greer and M. G. Ettlinger, 1949b. L-5-vinyl-2-thio-oxazolidone, an antithyroid compound from yellow turnip and from *Brassica* seeds. *J. Biol. Chem.* 181: 121-130.
- Barnes, M. McC., and A. A. Woodham, 1963. Prediction of quality in protein concentrates by laboratory procedures involving determination of soluble nitrogen. *J. Sci. Food Agr.* 14: 109-120.
- Bayley, H. S., D. C. Hill, J. D. Summers and S. J. Slinger, 1970. The value of rapeseed meal as a poultry feed. XIVth World's Poultry Congress abstracts, 1970, p. 221-222.
- Blakely, R. M., and R. W. Anderson, 1949. Studies with the rapeseed oilcake meal. I. The effect of various levels of rapeseed oilcake meal in the diet on the weight of the thyroid glands of turkey poults. *Sci. Agr.* 28: 393-397.
- Bolton, W., 1957. The digestibility by adult fowls of wheat fine middling, maize germ, maize gluten feed, soybean meal and groundnut meal. *J. Sci. Food Agr.* 8: 132-136.
- Briggs, G. M., and R. J. Lillie, 1946. Perosis caused by feeding high level of thiouracil. *Proc. Soc. Expt. Biol. Med.* 61: 430.

- Carew, L. B., Jr., and F. W. Hill, 1961. Effect of methionine deficiency on the utilization of energy by the chicks. *J. Nutrition*, 74: 185-190.
- Carpenter, K. J., and K. M. Clegg, 1956. The metabolizable energy of poultry feeding stuffs in relation to their chemical composition. *J. Sci. Food Agr.* 7: 45-51.
- Carroll, K. K., 1949. Isolation of an antithyroid compound from rapeseed (*Brassica napus*). *Proc. Soc. Exp. Biol. Med.* 71: 622-624.
- Clandinin, D. R., Ruth Renner and A. R. Robblee, 1959. Rapeseed oil meal studies. I. Effects of variety of rapeseed, growing environment and processing temperature on the nutritive value and chemical composition of rapeseed oil meal. *Poultry Sci.* 38: 1367-1372.
- Clandinin, D. R. and L. Bayly, 1960. Rapeseed oil meal studies. 2. Effects of feeding rapeseed oil meal on the structure of the thyroid glands of chickens. *Poultry Sci.* 39: 1239 (Abstract).
- Clandinin, D. R., and E. W. Tajcnar, 1961. Rapeseed oil meal studies. 3. Effects of variation in cooking and conditioning temperatures used during expeller processing of rapeseed on the fat and lysine content of rapeseed oil meals. *Poultry Sci.* 40: 291-293.
- Clandinin, D. R., L. Bayly and A. Caballero, 1966. Effects of ($\pm$)-5-vinyl-2-oxazolidinethione, A goitrogen in rapeseed meal on the rate of growth and thyroid function of chicks. *Poultry Sci.* 45: 833-838.
- Clandinin, D. R., 1967. Nutrient composition of expeller, prepress solvent, and solvent-processed rapeseed meals. *Poultry Sci.* 46: 1956-1957.
- Clandinin, D. R. and Jean Heard, 1968. Tannins in prepress solvent and solvent processed rapeseed meal. *Poultry Sci.* 47: 688.
- Clandinin, D. R., S. J. Slinger and J. M. Bell, 1972. Composition of Canadian rapeseed meal. p. 8-10. In *Canadian Rapeseed Meal in Poultry and Animal Feeding*. Rapeseed Association of Canada Publs. No. 15, Winnipeg.
- Conklin, R. L. and W. A. Maw, 1930. Pressure necrosis - a battery brooder feed problem. *Alabama Polytechnic Inst. Bul.*, 25: 35-37.

- Dow, D. S., and C. E. Allen, 1954. Rapeseed oilmeal in broiler rations with observations on the nature and control of its metabolic inhibitors. *Can. J. Agr.* 34: 607-613.
- Downey, R. K., 1972. Report given at Rapeseed Research Work Planning meeting held in Saskatoon, Saskatchewan.
- Drouliscos, N. J. and J. P. Bowland, 1969. Biological evaluation of rapeseed meal, soya-bean meal and casein fed to the weanling and the mature rat. *Br. J. Nutr.* 23: 113-123.
- Evans, R. J. and J. L. St. John, 1945. Estimation of the relative nutritive value of vegetable proteins by two chemical methods. *J. Nutrition*, 30: 209-217.
- Fink, K., 1944. The effect of the thyroid on jejunal secretion in the dog. *Am. J. Physiol.* 141: 598-605.
- Fraps, G. S., and E. C. Carlyle, 1942. Productive energy of some feeds as measured by gains of energy of growing chickens. *Texas Agr. Exp. Sta. Bull.* No. 625.
- Frölich, A., 1953. Experiments with different treatments of rapeseed meal. *Kungl. Lantbrukshogskolans Ann.* 20: 105-116.
- Gallup, W. D., and L. C. Norris, 1939. The amount of manganese required to prevent perosis in the chick. *Poultry Sci.* 18: 76-82.
- Gmelin, R., and A. J. Virtanen, 1959. A new type of enzymatic cleavage of mustard oil glucosides. *Acta. Chem. Scand.* 13: 1474-1475.
- Greer, M. A., 1956. Isolation from rustabage seed of progoitrin, the precursor of the naturally occurring antithyroid compound, goitrin (L-5-vinyl-2-thioxazolidone). *J. Am. Chem. Soc.* 78: 1260-1261.
- Greer, M. A., 1962. The isolation and identification of progoitrin from *Brassica* seed. *Arch. Biochem. Biophys.* 99: 369-371.
- Griesbach, W. E., 1941. Studies on experimental goitre. II. Changes in the anterior pituitary of the rat produced by the *Brassica* seed diet. *Brit. J. Exp. Path.* 22: 245-249.
- Griesbach, W. E., T. H. Kennedy and H. D. Purves, 1941. Studies on experimental goitre. III. The effect of goitrogenic diet on hypophysectomized rats. *Brit. J. Expt. Path.* 22: 249-254.

- Griesbach, W. E., and H. D. Purves, 1943. Studies on experimental goitre. V. Pituitary function in relation to goitrogenesis and thyroidectomy. Brit. J. Exp. Path. 24: 174-184.
- Hardinge, M. G., J. B. Swarner and H. Crooks, 1965. Carbohydrates in foods. J. Am. Diet. Assoc. 46: 197-204.
- Hercus, C. E., and H. D. Purves, 1936. Studies on endemic and experimental goitre. J. Hyg. Camb. 36: 183-203.
- Hill, F. W. and D. A. Anderson, 1958. Comparison of metabolizable energy and productive energy determinations with chicks. J. Nutrition, 64: 567-603.
- Hill, F. W., D. L. Anderson, R. Renner and L. B. Carew, Jr., 1960. Studies of the metabolizable energy of grains and grain products for chickens. Poultry Sci. 39: 573-579.
- Hill, F. W., 1965. Utilization of energy for growth by chicks. p. 327-332. In K. L. Blaxter, Energy Metabolism, Academic Press, London.
- Holmes, W. B. and R. Roberts, 1963. A perotic syndrome in chicks fed extracted rapeseed meal. Poultry Sci. 42: 803-809.
- Hrdlicka, J., H. Kozłowska, J. Pokorný and A. Rutkowski, 1964. Über rapsschrote. 3. Mitt. stickstoffhaltige substanzen der rapsschrote. Die Nahrung, 8(VI, VII): 537-543.
- Hrdlicka, J., H. Kozłowska, J. Pokorný and A. Rutkowski, 1965. Über rapsschrote. Mitt. saccharide in extraktionsschroten, Die Nahrung 9: 71-76.
- Jackson, M. L., 1958. Soil Chemical Analysis. p. 151-153. Prentice Hall Inc., Englewood Cliffs, N. J.
- Jull, M. A., 1951. Poultry Husbandry. p. 114. 3rd Ed. McGraw Hill Book Co. Inc., New York.
- Jurd, L. and Geissman, 1956. Absorption spectra of metal complexes of flavonoid compounds. J. Org. Chem. 27: 1395-1401.
- Karvanek, M., J. Pokorný, H. Kozłowska and A. Rutkowski, 1964. Über Rapsschrote. 5. Mitt. mineralstoffe. Die Nahrung. 8 (VIII): 675-680.

- Kennedy, T. H., and H. D. Purves, 1941. Studies on experimental goitre. I. The effect of *Brassica* seed diets on rats. Brit. J. Exp. Path. 22: 241-244.
- Kjaer, A., 1960. The Chemistry of Organic Natural Products, p. 122. Springer-Verlag, Vienna.
- Klain, G. J., D. C. Hill, H. D. Branion and J. A. Gray, 1956. The value of rapeseed oil meal and sunflower seed oil meal in chick starter rations. Poultry Sci. 35: 1315-1326.
- Kubota, D., and H. Morimoto, 1965. Nutritive value of feed-stuffs for poultry and reliability of digestible nutrients formula feeds calculated from the digestible crude protein and total digestible nutrients contents of ingredients (II). Japanese Poultry Sci. 2: 63-68.
- Lodhi, G.N., R. Renner and D. R. Clandinin, 1969. Studies on the metabolizable energy of rapeseed meal for growing chickens and laying hens. Poultry Sci. 48: 964-970.
- Lodhi, G. N., Ruth Renner and D. R. Clandinin, 1969. Available carbohydrate in rapeseed meal and soybean meal as determined by a chemical method and a chick bioassay. J. Nutrition, 99: 413-418.
- Lodhi, G. N., Ruth Renner and D. R. Clandinin, 1970. Factors affecting the metabolizable energy value of rapeseed meal. 2. Nitrogen absorbability. Poultry Sci. 49: 992-999.
- Lodhi, G. N., D. R. Clandinin and Ruth Renner, 1970. Factors affecting the metabolizable energy value of rapeseed meal. I. Goitrogens. Poultry Sci. 49: 289-294.
- Lodhi, G. N., 1970. Energy value of rapeseed meal for chickens Ph. D. thesis submitted to the University of Alberta, Edmonton.
- Lund, A. P. and W. M. Sandstrom, 1943. The proteins of various tree seeds. J. Agr. Res. 66: 349-356.
- Matsumoto, T., H. Itoh and Y. Akiba, 1968. Goitrogenic effects of (-)-5-vinyl-2-oxazolidinethione, a goitrogen in rapeseed, in growing chicks. Poultry Sci. 48: 1061-1069.
- Matterson, L. D., L. M. Potter, A. W. Arnold and E. P. Singsen, 1958. Studies in evaluating energy content of feed for the chicks. 2. Methods of evaluating feed ingredients for their metabolizable energy in the chick. Poultry Sci. 37: 1227 (Abstract).

- Olcott, H. S. and T. D. Fontaine, 1942. Effect of cooking on solubility of cottonseed meal proteins. *Industr. Eng. Chem.* 34: 714-716.
- Potter, L. M. and L. D. Matterson, 1960. Metabolizable energy of feed ingredients for growing chicks. *Poultry Sci.* 39: 781-782.
- Purves, H. D., 1943. Studies on experimental goitre. IV. The effect of di-iodotyrosine, tyrosine and thyroxine on the goitrogenic action of *Brassica* seed. *Brit. J. Exp. Path.* 24: 171-174.
- Rao, P. V. and D. R. Clandinin, 1970. Effect of method of determination on the metabolizable energy value of rapeseed meal. *Poultry Sci.* 49: 1069-1074.
- Rao, P. V., 1970, Personal Communication.
- Renner, R. and F. W. Hill, 1960b. Utilization of corn oil, lard and tallow by chickens of various ages. *Poultry Sci.* 39: 849-854.
- Russell, R. A. and E. S. Nasset, 1953. Influence of the thyroid gland on jejunal secretion in the dog. *Am. J. Physiol.* 172: 100-106.
- Scow, R. O. and V. G. Foglia, 1951. Effect of neonatal thyroidectomy, age and sex on intestinal absorption and tolerance of orally administered glucose in the rats. *Am. J. Physiol.* 166: 541-549.
- Sell, J. L., 1966. Metabolizable energy of rapeseed meal for the laying hens. *Poultry Sci.* 45: 854-856.
- Sibbald, I. R., J. D. Summers and S. J. Slinger, 1960. Factors affecting the metabolizable energy content of Poultry feed. *Poultry Sci.* 39: 544-556.
- Sibbald, I. R., S. J. Slinger and G. C. Ashton, 1962. Factors affecting the metabolizable energy content of poultry feeds. 5. The level of protein and of test material in the diet. 6. A note on the relationship between the digestible and metabolizable energy values. *Poultry Sci.* 41: 107-116.
- Sibbald, I. R., and S. J. Slinger, 1963a. A biological assay for metabolizable energy in poultry feed ingredients together with findings which demonstrate some of the problems associated with the evaluation of fats. *Poultry Sci.* 42: 313-325.

- Steel, R. G. D. and J. H. Torrie, 1960. Principles and Procedures of Statistics. McGraw-Hill Book Company, Inc., New York.
- Sun, D. C. H., H. Shay, H. Siplet and M. Gruenstein, 1954. Effect of experimental athyroidism and hypothyroidism upon gastric secretion in the rat. *Gastroenterology*, 27: 189-200.
- Turner, C. W. 1946. Effect of rapeseed on the thyroid of the chick. *Poultry Sci.* 25: 186-187.
- Van Etten, C. H., M. E. Daxenbichler and I. A. Wolff, 1969. Natural glucosinolates (thioglucosides) in foods and feeds. *J. Agr. Food Chem.* 17: 483-491.
- Vogel, A. I. 1961. A Text Book of Quantitative Inorganic Analysis Including Elementary Instrumental Analysis. p. 436-438. Longmans, Green and Co. Ltd., London.
- Vohra, P., F. H. Kratzer and M. A. Joslyn, 1966. The growth depressing and toxic effects of tannins to chicks. *Poultry Sci.* 45: 135-142.
- Wetter, L. R., 1955. The determination of mustard oils in rapeseed meal. *Can. J. Biochem. Physiol.* 33: 980-984.
- Wetter, L. R., 1957. The estimation of substituted thio-oxazolidones in the rapeseed meals. *Can. J. Biochem. Physiol.* 35: 293-297.
- Wetter, L. R. and B. M. Craig, 1959. Varietal and environmental effects on rapeseed isothiocyanate and thiooxazolidone content. *Can. J. Plant. Sci.* 39: 395-399.
- Wilgus, H. S., Jr., L. C. Norris and G. F. Heuser, 1936. The role of certain inorganic elements in the cause and prevention of perosis. *Science*, 84: 252-253.
- Witz, W. M., M. M. Carpenter and J. W. Hayward, 1950. Nutritional studies with rapeseed meal. *Poultry Sci.* 29: 786 (Abstract).
- Yapar, Z. and D. R. Clandinin, 1972. Effect of tannins in rapeseed meal on its nutritional value for chicks. *Poultry Sci.* 51: 222-228.
- Youngs, C. G., 1965. Processing of rapeseed meal. Rapeseed meal for livestock and Poultry, A Review. p. 24-31. Canada Department of Agri. Publs. no. 1257, Ottawa.

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